

Amendments to the Claims:

This listing of claims replaces all prior versions and listings of claims in the application:

Listing of Claims:

1 – 10. (Cancelled)

11. (Currently amended) A method of evaluating the deacetylation of a histone protein ~~or an N-terminal fragment thereof~~ in the presence of: (i) a Sir2 protein, (ii) NAD or an NAD-like compound, and (iii) an agent, the method comprising:

a) combining:

(i) a histone protein ~~or an N-terminal fragment thereof~~ that comprises an acetylated amino acid side chain,

(ii) an isolated or recombinantly produced Sir2 protein,

(iii) NAD or an NAD-like compound, and

(iv) an agent to be tested,

thereby producing a combination; and

b) determining if the histone protein ~~or the N-terminal fragment thereof~~ is deacetylated.

12 – 168. (Cancelled)

169. (Previously presented) The method of claim 11 wherein step (b) comprises electron-spray mass spectroscopy.

170. (Currently amended) The method of claim 11 further comprising comparing deacetylation of the histone protein ~~or the N-terminal fragment thereof~~ in the presence of the agent to deacetylation of the histone protein ~~or the N-terminal fragment thereof~~ in the absence of

the agent, wherein a difference in histone protein ~~or the N-terminal fragment thereof~~ deacetylation indicates that the agent alters Sir2 protein deacetylase activity.

171. (Previously presented) The method of claim 11 wherein the Sir2 protein is a human Sir2 protein.

172. (Previously presented) The method of claim 11 wherein the Sir2 protein is a murine Sir2 protein.

173. (Previously presented) The method of claim 11 wherein the Sir2 protein is a fusion protein.

174. (Currently amended) A method of evaluating the deacetylation of ~~The method of claim 11 wherein (i) in step (a) is the an N-terminal fragment tail of a an H3 or an H4 histone protein in the presence of: (i) a Sir2 protein, (ii) NAD or an NAD-like compound, and (iii) an agent, the method comprising:~~

a) combining:

(i) an N-terminal tail of an H3 or an H4 histone protein that comprises an acetylated amino acid side chain,

(ii) an isolated or recombinantly produced Sir2 protein,

(iii) NAD or an NAD-like compound, and

(iv) an agent to be tested,

thereby producing a combination; and

b) determining if the N-terminal tail of an H3 or H4 histone protein is deacetylated.

175. (Previously presented) The method of claim 11 wherein the histone protein is histone H3.

176. (Cancelled)

177. (Cancelled)

178. (Previously presented) The method of claim 11 wherein the histone protein is selected from the group consisting of an H2B, H3 and H4 histone protein.

179. (Previously presented) The method of claim 11 wherein the histone protein is acetylated on a lysine amino acid residue.

180. (Cancelled)

181. (Previously presented) The method of claim 11 wherein the acetylated amino acid is an acetylated lysine amino acid.

182. (Previously presented) The method of claim 11 wherein the Sir2 protein is an isolated Sir2 protein.

183. (Previously presented) The method of claim 11 wherein the Sir2 protein is a recombinantly produced Sir2 protein.

184. (Previously presented) The method of claim 11 wherein the combination comprises  $\text{MgCl}_2$ .

185. (Previously presented) The method of claim 11 wherein the combination comprises dithiothreitol (DTT).

186. (Previously presented) The method of claim 11 further comprising formulating the agent with a pharmaceutically acceptable carrier to provide a pharmaceutical composition.

187. (Previously presented) The method of claim 186 wherein the pharmaceutically acceptable carrier comprises a carbohydrate.

188. (Previously presented) The method of claim 11 wherein the combination comprises NAD.

189. (Previously presented) The method of claim 11 wherein the Sir2 protein is a Sir2 $\alpha$  protein.

190. (Previously presented) The method of claim 189 wherein the Sir2 $\alpha$  protein comprises SEQ ID NO:12.

191. (Currently amended) A method of evaluating the deacetylation of a histone protein or an N-terminal fragment of an H3 or an H4 histone protein thereof in the presence of a human Sir2 protein, NAD, and an agent, the method comprising:

a) providing a mixture comprising a histone protein or an N-terminal fragment of an H3 or an H4 histone protein thereof that comprises an acetylated amino acid side chain, an isolated or recombinantly produced human Sir2 protein, NAD, and an agent to be tested; and

b) determining if the histone protein or the N-terminal fragment of an H3 or an H4 histone protein thereof is deacetylated.

192. (Previously presented) The method of claim 191 wherein the mixture comprises MgCl<sub>2</sub>.

193. (Previously presented) The method of claim 191 wherein the mixture comprises dithiothreitol (DTT).

194. (Previously presented) The method of claim 11 or 191 wherein the Sir2 protein is produced in *E. coli*.

195. (Previously presented) The method of claim 11 or 191 wherein the agent is a protein.

196. (Previously presented) The method of claim 11 or 191 wherein the agent is a peptide.

197. (Previously presented) The method of claim 11 or 191 wherein the agent is naturally occurring.

198. (Previously presented) The method of claim 11 or 191 wherein the agent is non-naturally occurring.

199. (Previously presented) The method of claim 11 or 191 wherein the agent is chemically synthesized.

200. (Previously presented) The method of claim 11 or 191 wherein the agent is a carbohydrate.

201. (Previously presented) The method of claim 11 or 191 wherein the agent is a steroid.

202. (Previously presented) The method of claim 11 or 191 wherein the agent is a lipid.

203. (Previously presented) The method of claim 11 or 191 wherein the agent is an anion.

204. (Previously presented) The method of claim 11 or 191 wherein the agent is a cation.

205. (Previously presented) The method of claim 11 or 191 wherein the agent is an oligonucleotide.

206. (Previously presented) The method of claim 195 wherein the agent is an antibody.

207. (Previously presented) The method of claim 191 wherein the Sir2 protein is an isolated Sir2 protein.

208. (Previously presented) The method of claim 191 wherein the Sir2 protein is a recombinantly produced Sir2 protein.

209- 219. (Cancelled)

220. (New) The method of claim 174, wherein the fragment of the H3 histone protein comprises a sequence with SEQ ID NO:6.

221. (New) The method of claim 174, wherein the fragment of the H3 histone protein comprises amino acids 1-20 of a sequence with SEQ ID NO:6.

222. (New) The method of claim 174, wherein the fragment of the H4 histone protein comprises a sequence with SEQ ID NO:7 or SEQ ID NO:8.

223. (New) The method of claim 174, wherein the fragment of the H3 histone protein consists of a sequence with SEQ ID NO:6.

224. (New) The method of claim 174, wherein the fragment of the H3 histone protein consists of amino acids 1-20 of a sequence with SEQ ID NO:6.

225. (New) The method of claim 174, wherein the fragment of the H4 histone protein consists of a sequence with SEQ ID NO:7 or SEQ ID NO:8.

226. (New) The method of claim 191, wherein the fragment of the H3 histone protein comprises a sequence with SEQ ID NO:6.

227. (New) The method of claim 191, wherein the fragment of the H3 histone protein comprises amino acids 1-20 of a sequence with SEQ ID NO:6.

228. (New) The method of claim 191, wherein the fragment of the H4 histone protein comprises a sequence with SEQ ID NO:7 or SEQ ID NO:8.

229. (New) The method of claim 191, wherein the fragment of the H3 histone protein consists of a sequence with SEQ ID NO:6.

230. (New) The method of claim 191, wherein the fragment of the H3 histone protein consists of amino acids 1-20 of a sequence with SEQ ID NO:6.

231. (New) The method of claim 191, wherein the fragment of the H4 histone protein consists of a sequence with SEQ ID NO:7 or SEQ ID NO:8.